

PATENT  
09/888,309  
Docket 090/002

CLAIM AMENDMENTS

1 to 22. *(Cancelled)*

23. *(Previously presented)* A method for producing a neural cell population from human embryonic stem (hES) cells, comprising culturing progeny of the hES cells in a medium containing one or more added TGF- $\beta$  Superfamily Antagonists so as to produce a population in which at least 50% of the cells express either polysialylated NCAM or  $\beta$ -tubulin III.
24. *(Previously presented)* The method of claim 23, wherein the progeny are cultured in a medium containing noggin.
25. *(Previously presented)* The method of claim 23, wherein the progeny are cultured in a medium containing follistatin.
26. *(Previously presented)* The method of claim 23, wherein the medium further contains a neurotrophin.
27. *(Previously presented)* The method of claim 26, wherein the neurotrophin is NT-3 or BDNF.
28. *(Previously presented)* The method of claim 23, wherein the medium further contains a combination of factors selected from differentiation factors, neurotrophic factors, and survival factors.
29. *(Previously presented)* The method of claim 23, comprising differentiating the hES cells by plating them onto a solid surface without forming embryoid bodies or cell aggregates.
30. *(Previously presented)* The method of claim 29, wherein the solid surface comprises fibronectin or a polycation.
31. *(Previously presented)* The method of claim 23, wherein at least 10% of the MAP-2 positive cells in the produced population express tyrosine hydroxylase.
32. *(Previously presented)* The method of claim 23, further comprising combining the cell population with a compound, determining any phenotypic or metabolic changes in the cell that result from contact with the compound, and correlating the change with cellular toxicity or modulation caused by the compound.

PATENT  
09/888,309  
Docket 090/002

33. *(Previously presented)* The method of claim 23, further comprising identifying an mRNA expressed at a different level in the neural cell population, relative to the level in undifferentiated hES cells; and preparing a polynucleotide comprising a nucleotide sequence of at least 30 consecutive nucleotides contained in the identified mRNA.
- 
34. *(Currently amended)* ~~A set of two cultured cell populations, consisting of~~  
A system for producing differentiated cells from human embryonic stem (hES) cells, comprising:  
a first cell population comprising ~~undifferentiated cells from a line of human embryonic stem (hES)~~ the undifferentiated hES cells; and  
a second cell population, comprising progeny of the hES cells in a medium containing one or more added TGF- $\beta$  Superfamily Antagonists.
35. *(Currently amended)* ~~A set of two cultured cell populations, consisting of~~  
A system for producing differentiated cells from human embryonic stem (hES) cells, comprising:  
a first cell population comprising ~~undifferentiated cells from a line of human embryonic stem (hES)~~ the undifferentiated hES cells; and  
a second cell population, comprising at least ~10% hES derived neural cells, identifiable by the criteria that they are progeny of said hES ~~cell line~~ cells and express both MAP-2 and tyrosine hydroxylase.
36. *(Currently amended)* ~~The set of cell populations system~~ of claim 35, wherein the second population has been produced from cells of the first population (or their progeny) by the method of claim 23.
37. *(New)* The system of claim 36, wherein the progeny of the hES cells are cultured in a medium containing noggin.
38. *(New)* The system of claim 36, wherein the progeny of the hES cells are cultured in a medium containing follistatin.
39. *(New)* The system of claim 36, wherein the medium further contains a neurotrophin.
40. *(New)* The system of claim 39, wherein the neurotrophin is NT-3 or BDNF.
41. *(New)* The system of claim 34, wherein the second cell population is in a medium containing

PATENT  
09/888,309  
Docket 090/002

noggin.

42. (New) The system of claim 34, wherein the second cell population is in a medium containing follistatin.
43. (New) The system of claim 34, wherein the medium further contains a neurotrophin.
44. (New) The system of claim 43, wherein the neurotrophin is NT-3 or BDNF.
45. (New) The system of claim 34, wherein the second cell population has been obtained by a process comprising differentiating the hES cells by plating them onto a solid surface without forming embryoid bodies or cell aggregates.
46. (New) The system of claim 46, wherein the solid surface comprises fibronectin or a polycation.
47. (New) A method for testing a substance for its effect on differentiated cells, comprising combining the second cell population of claim 34 with the compound, determining any phenotypic or metabolic changes in the cell population that result from contact with the substance, and correlating the change with cellular toxicity or modulation caused by the substance.
48. (New) A method for preparing a polynucleotide containing a gene sequence that undergoes a change in expression level during differentiation of hES cells, comprising identifying an mRNA expressed at a different level in the second cell population of claim 34 relative to the level in the first cell population; and preparing a polynucleotide comprising a nucleotide sequence of at least 30 consecutive nucleotides contained in the identified mRNA.